PCT WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification [6] : **A61K 31/28, 33/24**	**A1**	**(11) International Publication Number:** **WO 95/05814** **(43) International Publication Date:** 2 March 1995 (02.03.95)

(21) International Application Number: PCT/GB94/01817

(22) International Filing Date: 19 August 1994 (19.08.94)

(30) Priority Data:
9317686.5 25 August 1993 (25.08.93) GB

(71) Applicant *(for all designated States except US)*: JOHNSON MATTHEY PUBLIC LIMITED COMPANY [GB/GB]; 78 Hatton Garden, London EC1N 8JP (GB).

(72) Inventors; and
(75) Inventors/Applicants *(for US only)*: ABRAMS, Michael, Jeffrey [US/US]; Creek Road, RD No. 2, Box 31, Glenmore, PA 19343 (US). FRICKER, Simon, Paul [GB/GB]; 30 Glebe Road, Sonning-on-Thames, Reading RG4 0XH (GB). MURRER, Barry, Anthony [GB/GB]; 17 Carnarvon Road, Reading RG1 5SB (GB). VAUGHAN, Owen, John [GB/SE]; Folvagen 14, S-152 57 Ostertalie (SE).

(74) Agent: WISHART, Ian, Carmichael; Johnson Matthey Technology Centre, Blounts Court, Sonning Common, Reading RG4 9NH (GB).

(81) Designated States: AM, AT, AU, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, ES, FI, GB, GE, HU, JP, KE, KG, KP, KR, KZ, LK, LT, LU, LV, MD, MG, MN, MW, NL, NO, NZ, PL, PT, RO, RU, SD, SE, SI, SK, TJ, TT, UA, US, UZ, VN, European patent (AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG), ARIPO patent (KE, MW, SD).

Published
With international search report.

(54) Title: PHARMACEUTICAL COMPOSITIONS COMPRISING METAL COMPLEXES

(57) Abstract

New pharmaceutical compositions and pharmaceutical compositions comprising metal complexes have activity against diseases caused by or related to overproduction or localised high concentration of nitric oxide in the body.

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AT	Austria	**GB**	United Kingdom	**MR**	Mauritania
AU	Australia	**GE**	Georgia	**MW**	Malawi
BB	Barbados	**GN**	Guinea	**NE**	Niger
BE	Belgium	**GR**	Greece	**NL**	Netherlands
BF	Burkina Faso	**HU**	Hungary	**NO**	Norway
BG	Bulgaria	**IE**	Ireland	**NZ**	New Zealand
BJ	Benin	**IT**	Italy	**PL**	Poland
BR	Brazil	**JP**	Japan	**PT**	Portugal
BY	Belarus	**KE**	Kenya	**RO**	Romania
CA	Canada	**KG**	Kyrgystan	**RU**	Russian Federation
CF	Central African Republic	**KP**	Democratic People's Republic of Korea	**SD**	Sudan
CG	Congo			**SE**	Sweden
CH	Switzerland	**KR**	Republic of Korea	**SI**	Slovenia
CI	Côte d'Ivoire	**KZ**	Kazakhstan	**SK**	Slovakia
CM	Cameroon	**LI**	Liechtenstein	**SN**	Senegal
CN	China	**LK**	Sri Lanka	**TD**	Chad
CS	Czechoslovakia	**LU**	Luxembourg	**TG**	Togo
CZ	Czech Republic	**LV**	Latvia	**TJ**	Tajikistan
DE	Germany	**MC**	Monaco	**TT**	Trinidad and Tobago
DK	Denmark	**MD**	Republic of Moldova	**UA**	Ukraine
ES	Spain	**MG**	Madagascar	**US**	United States of America
FI	Finland	**ML**	Mali	**UZ**	Uzbekistan
FR	France	**MN**	Mongolia	**VN**	Viet Nam
GA	Gabon				

PHARMACEUTICAL COMPOSITIONS COMPRISING METAL COMPLEXES

This invention relates to new pharmaceutical compositions and to pharmaceutical compositions having activity against diseases caused by or related to overproduction or localised high concentration of nitric oxide in the body.

Nitric oxide (NO) plays a varied and vital role in the human body. For example, NO plays a vital role in the control of blood pressure; it acts as a neurotransmitter; it plays a role in inhibition of platelet aggregation (important in thrombosis or blockages of the blood vessels), and in cytostasis (important in

fighting of tumours). Overproduction of NO however, has been implicated in a number of disease states, including vascular/pressor diseases such as septic shock, post-ischaemic cerebral damage, migraine, and dialysis induced renal hypotension; immunopathologic diseases such as hepatic damage in inflammation and sepsis, allograft rejection, graft versus host diseases, diabetes and wound healing; neurodegenerative diseases such as cerebral ischaemia, trauma, chronic epilepsy, Alzheimer's disease, Huntington's disease, and AIDS dementia complex; and side effects of treatment such as restenosis following angioplastic treatment and secondary hypotension following cytokine therapy.

Pharmacological modulation of nitric oxide in any of these disease states should prove extremely beneficial.

One above-mentioned disease relating to overproduction of NO, is septic shock. This is precipitated by local septicaemia or endotoxaemia, (high local levels of bacterial endotoxins). The result is activation of macrophages, lymphocytes, endothelial cells and other cell types capable of producing NO, further mediated by cytokine production by these cells. The activated macrophages produce excess NO which causes vasodilation of the blood vessels, and results in local vascular damage and vascular collapse. This destruction of vascular integrity may be so great that it leads to the collapse of haemodynamic homeostasis, the end result being death.

Current ideas for pharmacological modulation of nitric oxide in such diseases are based on dealing with the mediators of septic shock, such as cytokines, endotoxins, and platelet activating factor (PAF). The approaches include use of antibodies to cytokines such as tumour necrosis factor (TNF), receptor antagonists such as interleukin 1, (IL-1), antibodies to lipopolysaccharide (the endotoxin produced by gram negative bacteria, and PAF antagonists. All such approaches while challenging a factor mediating septic shock, do not attempt to deal with the aetiology, or cause, of the disease. Recent advances in understanding of NO have lead to the proposal that inhibitors of the NO synthase enzyme, such as N^G-monomethy-L-arginine (L-NMMA), may be useful in the treatment of septic shock and other NO overproduction related to diseases since they inhibit NO production. While these inhibitors have shown some utility in animal models and preliminary clinical studies, they have the disadvantage of undesirably inhibiting total NO synthesis in the body.

An aim of the present invention is to provide new and previously indicated pharmaceutical compositions which are able to modulate NO levels in the body by scavenging, or removing, NO in situ so that necessary NO synthesis continues while dangerous excesses are removed. We have found that certain metal complexes have the ability to carry out this important role.

Some metal complexes are known in pharmaceutical compositions for the treatment of diseases of the human body. For example, certain complexes of platinum and ruthenium have been used or indicated in the treatment of cancer.

Metal complexes have not however been previously indicated in the treatment of NO overproduction related diseases.

This invention provides for the use of a neutral, anionic or cationic metal complex having at least one site for coordination with NO, of formula

$$[M_a(X_bL)_cY_dZ_e]^{n\pm} \qquad \text{formula I,}$$

in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease,

where: M is a metal ion or a mixture of metal ions;

X is a cation or a mixture of cations;

L is a ligand, or mixture of ligands each containing at least two different donor atoms selected from the elements of Group IV, Group V or Group VI of the Periodic Table;

Y is a ligand, or a mixture of the same or different ligands each containing at least one donor atom or more than one donor atom, which donor atom is selected from the elements of Group IV, Group V or Group VI of the Periodic Table;

and Z is a halide or pseudohalide ion or a mixture of halide ions and pseudohalide ions;

a = 1-3; b = 0-12; c = 0-18; d = 0-18; e = 0-18; and n = 0-10;

provided that at least one of c, d and e is 1 or more;

and where c is 0; b is also 0;

and where a is 1; c, d and e are not greater than 9;

and where a is 2; c, d and e are not greater than 12.

By "complex" in this specification is meant a neutral complex or anionic or cationic species.

The term "Group" which is used herein is to be understood as a vertical column of the periodic table in which elements of each Group have similar physical and chemical properties. The definition of the Periodic Table is that credited to Mendeleev; Chambers Dictionary of Science and Technology, 1974. Published by W & R Chambers Ltd.

This invention may also be stated as providing a method of attenuation of NO levels where NO is implicated in diseases of the human body, comprising administering a pharmaceutical composition containing a neutral, anionic or cationic metal complex of formula I.

This invention may also provide for the use of a neutral, anionic or cationic metal complex of formula I in the manufacture of a medicament for the treatment of NO overproduction related disease.

This invention may also be stated as providing a method of treatment of diseases of the human body resultant of overproduction of NO in the human body, comprising administering a pharmaceutical composition containing a neutral, anionic or cationic metal complex of formula I.

Where the formula I represents an anionic species, a cation will also be present. Where formula I represents a cationic species, an anion will also be present. The metal complexes may be hydrated.

Preferably, M is a first, second or third row transition metal ion. For example, M may be an Rh, Ru, Os, Mn, Co, Cr or Re ion, and is preferably an Rh, Ru or Os ion.

Suitably M is in an oxidation state III. We have found surprisingly that when the metal ion for example ruthenium is in oxidation state III, the rate at which it binds with NO is significantly faster than when it is in oxidation state II.

X may be any cation, such as a mono-, di- or tri-valent cation. Suitable cations may be H^+, K^+, Na^+, NH_4^+ or Ca^{2+}. Conveniently X may be H^+, K^+ or Na^+.

Preferably, L is a ligand containing both nitrogen and oxygen donor atoms. Examples of suitable such ligands include ethylenediamine -N,N'-diacetic acid (edda), ethylenediaminetetraacetic acid (edta), nitrilotriacetic acid (nta),

dipicolinic acid (dipic), picolinic acid (pic), diethylenetriaminepentaacetic acid (dtpa), thiobis(ethylenenitrilo)tetraacetic acid (tedta), dithioethanebis(ethylene-nitrilo)tetraacetic acid (dtedta) and N-(2-hydroxyethyl)ethylenediamine-triacetic acid (hedtra).

Preferably, Y is a ligand containing nitrogen, oxygen, sulphur, carbon, or phosphorus donor groups. Suitable nitrogen donor groups may be for example ammine, amine, nitrile and nitride or derivations thereof. Suitable oxygen donor groups may be for example carboxylic acid, ester or derivations thereof, water, oxide, sulphoxide, hydroxide, acetate, lactate, propionate, oxalate and maltolate. Suitable sulphur donor groups may be for example sulphoxide, dialkylsulphide, dithiocarbamate or dithiophosphate. Suitable carbon donor groups may be for example carbon monoxide or isocyanide. Suitable phosphorus donor groups may be for example trialkylphosphine.

Z may be any halide and is preferably chloride, bromide or iodide. Most conveniently, Z is chloride.

Examples of metal complexes for use according to the present invention include optionally hydrated ruthenium complexes of formula

$$[Ru(H_{0-6}L'')_{1-3}Y_{0-2}Cl_{0-4}]^{(0-4)\pm} \qquad \text{formula II,}$$

where L'' is an amide or ester or derivative thereof, or a polydentate aminocarboxylate ligand, for example edta, nta, dipic, pic, edda, tropolone, dtpa, hedtra, tedta or dtedta or diamide of edta or dtpa or a mixture of any of these, and Y is as defined above and may for example be selected from acetylacetone (acac), a β-diketonate, water, dimethylsulphoxide (dmso), carboxylate, bidentate carboxylate, catechol, kojic acid, maltol, hydroxide, tropolone, malonic acid, oxalic acid, 2,3-dihydroxynaphthalene, squaric acid, acetate, a sulphate and a glycolate. The skilled addressee will be able to substitute other known ligands at Y and which will fall within the scope of the inventions. Preparative methods of tedta, dtedta and diamide of edta and dtpa are described in the following references respectively:

P Tse & JE Powell, Inorg Chem, (1985), 24, 2727

G Schwartzenbach, H Senner, G Anderegg, Helv Chim Acta 1957, 40, 1886

MS Konings, WC Dow, DB Love, KN Raymond, SC Quay and SM Rocklage, Inorg Chem (1990), 29, 1488-1491

PN Turowski, SJ Rodgers, RC Scarrow and KN Raymond, Inorg Chem (1988), 27, 474-481.

Where the complex of formula II is an anion, a cation will be required. For example the complexes of formula II are present in

$K[Ru(Hedta)Cl]2H_2O$

$[Ru(H_2edta)(acac)]$

$K[Ru(hedtra)Cl]H_2O$

$K[Ru(dipic)_2]H_2O$

$(H_2pic)[RuCl_2(pic)_2](Hpic)H_2O$

$K[Ru(H_2edta)Cl_2]H_2O$

$K[Ru(Hnta)_2]½H_2O$

$K[Ru(H_2dtpa)Cl]H_2O$

$[Ru(Hhedtra)acac]H_2O$

$[Ru(Hhedtra)trop]$

$[Ru(H_3dtpa)Cl]$

Complexes of formula II have not to the best of our knowledge been previously indicated in any pharmaceutical composition. Therefore the present invention also provides a pharmaceutical composition containing an optionally hydrated ruthenium complex of formula II.

Further examples of metal complexes for use according to the present invention include optionally hydrated complexes of formula III

$$[M_{1-3}Y_{1-18}Cl_{0-18}]^{(0-6)\pm} \qquad \text{formula III}$$

Where Y is a sulphur donor ligand. For example, such complex is present in

$[Ru(mtc)_3]$ (mtc = 4-morpholinecarbodithoic acid)

$Ru(S_2CNCH_2CH_2NMeCH_2CH_2)_3½H_2O$

Complexes of formula III in which Y is a sulphur donor ligand have not to the best of our knowledge been previously indicated in any pharmaceutical composition. Therefore, the present invention also provides a pharmaceutical composition containing an optionally hydrated complex of formula III wherein Y is a sulphur donor ligand.

Yet further examples of metal complexes for use according to the present invention include optionally hydrated complexes of ruthenium of formula

$$[M'''_{1-3}Y'''_{1-18}Cl_{0-18}]^{(0-6)\pm} \qquad \text{formula III}$$

where M''' is ruthenium and Y''' is an oxygen-donor ligand such as acetate, lactate, water, oxide, propionate (COEt), oxalate (ox), or maltolate (maltol) or a combination of these. For example complexes of formula III are present in

$[Ru_3O(OAc)_6](OAc)$

$[Ru_3O(lac)_6](lac)$

$[Ru_2(OAc)_4]NO_3$

$[Ru_2(OCOEt)_4]NO_3$

$K_3[Ru(ox)_3]$

$[Ru_2(OAc)_4]Cl$

$[Ru(maltol)_3]$

Some complexes of formula III have not to the best of our knowledge been previously indicated in any pharmaceutical composition. Therefore the present invention also provides a pharmaceutical composition containing an optionally hydrated complex of ruthenium of formula III wherein M''' is ruthenium and Y''' is an oxygen-donor ligand selected from the group acetate, lactate, oxide, propionate and maltolate.

Further examples of metal complexes for use according to the present invention include optionally hydrated complexes of ruthenium of formula

$$[RuY^{IV}_{1-9}Cl_{1-9}]^{(0-4)\pm} \qquad \text{formula IV}$$

where Y^{IV} is a nitrogen-donor ligand such as ammine, ethylenediamine (en), pyridine (py), 1,10-phenanthroline (phen), 2,2'-bipyridine (bipy) or 1,4,8,11-tetra-azacyclotetradecane (cyclam), 2,3,7,8,12,13,17,18-octaethylporphyrin (oep) or a combination of these. For example complexes of formula IV are present in

$[Ru(NH_3)_5Cl]Cl_2$

$[Ru(en)_3]I_3$

trans-$[RuCl_2(py)_4]$

$K[Ru(phen)Cl_4]$

$[Ru(cyclam)Cl_2]Cl$

$K[Ru(bipy)Cl_4]$

$[Ru(NH_3)_6]Cl_3$

$[Ru(NH_3)_4Cl_2]Cl$

Ru(oep)Ph

Some complexes of formula IV have not to the best of our knowledge been previously indicated in any pharmaceutical composition. Therefore the present invention also provides a pharmaceutical composition containing an optionally hydrated complex of ruthenium of formula IV wherein Y^{IV} is a nitrogen-donor ligand selected from the group en, py, phen, bipy, cyclam and oep. Derivations of these ligands can be prepared by a skilled addressee and which will fall within the scope of the inventions.

Still further examples of metal complexes for use according to the present invention include optionally hydrated complexes of ruthenium or osmium of general formula

$$[M_{1-3}Y^{V}_{1-18}Cl_{0-18}]^{(0-6)\pm} \qquad \text{formula V}$$

where Y^V is a combination of donor ligands such as are described hereinabove, for example ammine, dmso, oxalate, bipy, acac and MeCN. Complexes of formula V are present in for example

$[Ru(NH_3)(dmso)_2Cl_3]$

cis-$[Ru(dmso)_4Cl_2]$

cis-$[Ru(NH_3)(dmso)_3Cl_2]$

$[Ru(dmso)_3Cl_3]$

$[Os(ox)(bipy)_2]H_2O$

$[Ru(acac)_2(MeCN)_2]CF_3SO_3$

The complex ions of the latter two compounds above have not to the best of our knowledge been previously indicated in any pharmaceutical composition. Therefore the present invention also provides a pharmaceutical composition containing an optionally hydrated complex of formula $[Os(ox)(bipy)_2]$; and further a pharmaceutical composition containing an optionally hydrated complex of formula $[Ru(acac)_2(MeCN)_2]^+$.

In use the complexes of the present invention may be included as an active component in a pharmaceutical composition containing an optionally hydrated complex of any of formulae I-V, in admixture with a pharmaceutically acceptable carrier or diluent. Said pharmaceutical composition may be formulated according to well known principles, and may be in the form of a solution or suspension for parenteral administration in single or repeat doses or be in capsule, tablet, dragee, or other solid composition or as a solution or suspension for oral administration, or formulated into pessaries or suppositories, or sustained release forms of any of the above. The solution or suspension may be administered by a single or repeat bolus injection or continuous infusion, or any other desired schedule. Suitable diluents, carriers, excipients and other components are known. Said pharmaceutical composition may contain dosages determined in accordance with conventional pharmacological methods, suitable to provide active complexes

in the dosage range in humans of 1mg to 10g per day. Actual required dosage is largely dependent on where in the body there is the excess concentration of NO and for how long overproduction continues or attenuation of NO levels, where NO is implicated in disease, is required.

This invention will now be illustrated by Example.

A number of commercially available compounds, and compounds prepared by routes known in the literature, containing the complexes of the present invention were tested *in vitro*, *in vitro* cell culture, and *ex-vivo* in order to determine ability to coordinate with NO. The complexes tested were as follows:

Example	Compound	Literature Reference for Preparation
1	$K[Ru(Hedta)Cl]2H_2O$	AA Diamantis & JV Dubrawski, Inorg.Chem.,(1981),20,1142-50
2	$[Ru(H_2edta)(acac)]$	AA Diamantis & JV Dubrawski, Inorg.Chem.,(1983),22,1934-36
3	$K[Ru(hedtra)Cl]H_2O$	HC Bajaj & R van Eldik, Inorg.Chem.(1982),28,1980-3
4	$K[Ru(dipic)_2]H_2O$	NH Williams & JK Yandell, Aust.J.Chem.(1983),36(12),2377-2386
5	$(H_2pic)[RuCl_2(pic)_2](Hpic)H_2O$	JD Gilbert, D Rose & G Wilkinson, J.Chem.Soc.(A),(1970),2765-9

6	$K[Ru(H_2edta)Cl_2]H_2O$	AA Diamantis & JV Dubrawski, Inorg.Chem.(1981),20,1142-50
7	$K[Ru(Hnta)_2]½H_2O$	MM Taqui Khan, A Kumar & Z Shirin, J. Chem. Research (M), (1986),1001-1009
8	$K[Ru(H_2dtpa)Cl]H_2O$	MM Taqui Khan, A Kumar & Z Shirin, J. Chem. Research (M). (1986), 1001-1009
9	$[Ru_3O(lac)_6](lac)$	A Spencer & G Wilkinson, J. Chem. Soc. Dalton Trans, (1972), 1570-77
10	$[Ru_3O(OAc)_6](OAc)$	A Spencer & G Wilkinson, J.Chem. Soc. Dalton Trans. (1972), 1570-77
11	$[Ru_2(OAc)_4]NO_3$	M Mukaida, T Nomura & T Ishimori, Bull. Chem. Soc. Japan, (1972) ,45, 2143-7
12	$[Ru_2(OCOEt)_4]NO_3$	A Bino, FA Cotton & TR Felthouse, Inorg. Chem. (1979), 18, 2599-2604
13	$K_3[Ru(ox)_3]$	CM Che, SS Kwong, CK Poon, TF Lai & TCW Mak Inorg. Chem. (1985), 24, 1359-63

14	$[Ru_2(OAc)_4]Cl$	RW Mitchell, A Spencer & G Wilkinson J. Chem. Soc. Dalton Trans., (1973), 846-54
15	$[Ru(NH_3)_5Cl]Cl_2$	AD Allen, F Bottomley, RO Harris, VP Reinsalu & CV Senoff J. Amer. Chem. Soc. (1967), 89, 5595-5599
16	$[Ru(en)_3]I_3$	TJ Meyer & H Taube Inorg. Chem. (1968), 7, 2369-2379
17	$K[RuCl_4(phen)]H_2O$	BR James & RS McMillan Inorg. Nucl. Chem. Lett. (1975), 11(12) 837-9
18	$[Ru(cyclam)Cl_2]Cl$	PK Chan, DA Isabirye & CK Poon Inorg. Chem. (1975), 14, 2579-80
19	$K[RuCl_4(bipy)]$	BR James & RS McMillan Inorg. Nucl. Chem. Lett. (1975), 11(12), 837-9
20	$[RuCl_3(dmso)_2(NH_3)]$	Patent: International Publication No WO 91/13553
21	$[Ru(NH_3)_6]Cl_3$	Matthey Catalogue Sales: Cat No [190245]

22	cis-$[RuCl_2(dmso)_4]$	EA Alessio, G Mestroni, G Nardin, WM Attia, M Calligaris, G Sava & S Zorget Inorg. Chem. (1988), 27, 4099-4106
23	cis-$[RuCl_2(dmso)_3(NH_3)]$	M Henn, E Alessio, G Mestroni, M Calligaris & WM Attia Inorg. Chim. Acta, (1991), 187, 39-50
24	$[RuCl_3(dmso)_3]$	E Alessio, G Balducci, M Calligaris, G Costa, WM Attia & G Mestroni Inorg. Chem. (1991), 30, 609-618
25	$[Ru(mtc)_3]$	AR Hendrickson, JM Hope & RL Martin J. Chem. Soc. Dalton Trans. (1976), 20, 2032-9
26	$[Ru(maltol)_3]$	WP Griffith & SJ Greaves Polyhedron, (1988), 7(19), 1973-9
27	$[Ru(acac)_2(MeCN)_2]CF_3SO_3$	Y Kasahara, T Hoshino, K Shimizu & GP Sato Chem. Lett. (1990),3,381-4
28	$K_2[RuCl_5(H_2O)]$	Matthey Catalogue Sales: Cat No [190094]

29	$[Os(ox)(bipy)_2].H_2O$	DA Buckingham, FP Dwyer, HA Goodwin & AM Sargeson Aust.J.Chem.(1964),325-336
		GM Bryant, JE Fergusson & HKJ Powell Aust.J.Chem.(1971), 24(2),257-73
30	$[Ru(NH_3)_4Cl_2]Cl$	SD Pell, MM Sherban, V Tramintano & MJ Clarke Inorg Synth, (1989), 26, 65.
31	[Ru(Hedtra)(dppm)]	MM Taqui Khan, K Venkatasubramanian, Z Shirin, MM Bhadbhade J Chem Soc Dalt Trans (1992), 885-890
32	Ru(oep)Ph	M Ke, SJ Rettig, BR James and D Dolphin J Chem Soc Chem Commun (1987), 1110

A number of new compounds were prepared according to the following protocols. The first four compounds are examples of ruthenium complexes of formula $[Ru(H_{0-6}L'')_{1-3}\ Y_{0-2}\ Cl_{0-4}]^{(0-4)\pm}$ (formula II), the subsequent two are examples of $[M_{1-3}Y_{1-8}Cl_{0-18}]^{(0-6)\pm}$ (formula III).

Preparation of [Ru(Hhedtra)acac].H_2O

Excess acetylacetone ($1cm^3$) was added to an aqueous solution ($5cm^3$) of K[Ru(hedtra)Cl] (0.5g). The solution colour changed to violet. The mixture was warmed for 20 minutes then left to stand at room temperature for 20 minutes. The violet solution was extracted with chloroform ($20cm^3$). The extraction was repeated twice more. A violet product precipitated from the aqueous fraction. The product was filtered, washed in acetone and dried in vacuo, yield 0.1g (18%).

Anal. Calc. for $C_{15}H_{25}O_{10}N_2Ru$: C, 36.43; H, 5.11; N, 5.70. Found: C, 36.16; H, 5.42; N, 5.61%.

Preparation of [Ru(Hhedtra)trop]$2H_2O$

A three-fold excess of tropolone (0.78g) dissolved in 50:50 water/absolute ethanol ($5cm^3$) was added to a warm aqueous solution of K[Ru(hedtra)Cl] ($10cm^3$). The mixture was heated for 1 hour. On cooling, the dark green mixture was extracted with 3 x $20cm^3$ portions of dichloromethane. On standing, a dark green product precipitated from the aqueous fraction. The product was filtered, washed with water ($1cm^3$), ether and dried in vacuo, yield 0.4g (36%). Anal. Calc. for $C_{17}H_{22}N_2O_9Ru.2H_2O$: C, 38.13; H, 4.86; N, 5.23. Found: C, 38.55; H, 4.67; N, 5.28%.

Preparation of [Ru(H_3dtpa)Cl]

$K_2[RuCl_5H_2O].xH_2O$ (1g) was suspended in $HClO_4$ ($15cm^3$, 1mM) and diethylenetriaminepentaacetic acid (1.05g) added. The reaction mixture was heated under reflux for 1.5 hours forming a yellow/brown solution. On cooling a yellow product crystallised which was collected by filtration, washed with 90% absolute ethanol/water, diethyl ether and dried in vacuo, yield 0.75g, 53%.

Anal. calcd. for $C_{14}H_{21}N_3O_{10}ClRu$: C, 31.85; H, 3.98; N, 7.96; Cl, 6.73. Found: C, 29.77; H, 3.81; N, 7.36; Cl, 6.64.

Preparation of K[RuHHBEDCl]3H_2O

0.41g of $K_2[RuCl_5]xH_2O$ was dissolved in water (20ml). To this solution was added 1 equivalent (0.39g) of N,N'di(2-hydroxy-benzyl)ethylene-diamine N,N-diacetic acid (hbed) dissolved in water (50ml) with KOH (0.12g) and MeOH (1ml). This mixture was heated at reflux for 90 minutes. Upon cooling a dark, insoluble precipitate formed. This material was removed by filtration and the resulting red-violet solution was taken to dryness by rotary evaporation. Trituration with water and washing with acetone yielded 90mg of a dark solid.

Anal. calcd. for $C_{18}H_{22}N_2O_9RuClK$: C, 36.89; H, 3.96; N, 4.78; Cl, 6.04. Found: C, 37.09; H, 4.23; N, 4.92; Cl, 6.28.

Preparation of $Ru(S_2CNCH_2CH_2NMeCH_2CH_2)_3½H_2O$

$Me_4N[S_2CNCH_2CH_2NMeCH_2CH_2]$ was made by the standard method and crystallised from methanol-ether in 71% yield.

$RuCl_3xH_2O$, 0.50g, 2.15mmol was refluxed in 30ml of methanol for 10 minutes and cooled. 1.87g, 7.50mmol of $Me_4N[S_2CNCH_2CH_2NMeCH_2CH_2]$ was added and the mixture refluxed for 16 hours. After cooling 0.72g of crude product was filtered off, dissolved in dichloromethane and filtered. The filtrate was loaded into 15cc of basic alumina and eluted with dichloromethane. Removal of solvent and crystallisation from dichloromethane with ether by vapour-phase diffusion gave 0.51g, 0.80mmol, 37% of brown-black crystals, $Ru(S_2CNCH_2CH_2NMeCH_2CH_2)_3½H_2O$.

Analysis for $C_{18}H_{34}N_6O_{.5}RuS_6$: Calc: C, 34.00; H, 5.39; N, 13.22; S, 30.25. Found: C, 34.21; H, 5.47; N, 13.12; S, 30.36.

Preparation of $Ru[S_2P(OC_2H_2OC_2H_4OMe)_2]_3$

$K[S_2P(OC_2H_4OC_2H_4OMe)_2]_3$was made by standard method and crystallised from methanol in 76% yield.

$RuCl_3xH_2O$, 1.00g, 4.30mmol was refluxed in 50ml of 0.1 N HCl with 1ml of ethanol for 20 minutes and cooled. To this solution was added 5.28g (excess) $K[S_2P(OC_2H_4OC_2H_4OMe)_2]$ and the mixture stirred at 30°C for 1 hour. the reaction mixture was extracted with dichloromethane and the solvent removed.

The residue was extracted with ether-hexane and solvents removed. This residue was crystallised from 25ml of hot ether by cooling to -20°C giving 2.98 of red crystals. 2.41g of the crude product was purified by chromatography on 60cc of silica gel with 5% ethanol in ether. The first band was collected, reduced to dryness and crystallised from ether by cooling to -20°C. The yield of red crystals, $Ru(S_2P\{OC_2H_4OC_2H_4OMe\}_2)_3$, was 2.16g, 56%.

Analysis for $C_{30}H_{66}O_{18}P_3RuS_6$: Calc: C, 32.72; H, 6.04; S, 17.47. Found: C, 32.68; H, 6.08; S, 17.16.

In the *in vitro* tests, which were carried out in an atmosphere of argon, each compound (1×10^{-4} moles) was dissolved in double-distilled deionized and deoxygenated water. The resulting solution was placed in a three-necked pear-shaped flask and stirred by a magnetic stirrer at constant speed of 1000rpm, at a constant temperature in the range 20°C-24°C. A manometer was attached to the flask, and purified, dried nitric oxide gas (known volume in the range 3-5cm^3) was introduced via a septum, using a gas syringe, at atmospheric pressure into the headspace above the reaction solution. The pressure within the flask was recorded periodically over a period of one hour.

A control experiment was carried out according to the above but without any complex present.

The recorded pressures in association with the results of the control experiment were analysed in order to determine the rate of NO uptake as a function of time for each compound tested.

On completion of each *in vitro* test, the reaction solution was freeze-dried. An infrared spectrum of the freeze-dried product provided information on metal-NO bond formation.

In the *in vitro* cell culture tests, murine (RAW264) macrophage cell lines, which can be induced to produce nitric oxide, were seeded, 10^6cells/well, onto 24 well culture plates of 2ml volume per well, in Eagles modified minimal essential medium (MEM) plus 10% foetal bovine serum without phenol red.

The cells were activated to produce nitric oxide, with 10µg/ml lipopolysaccharide and 100 units/ml interferon γ for 18 hours. Concurrently, test compounds made up in MEM were added at non-cytotoxic concentrations.

Control cells as above, which were activated to produce nitric oxide as above, but to which no test compound was added, were used as a measure of the amount of nitric oxide produced by the cells during the tests.

Background nitric oxide was assessed by measurement of nitrate and nitrite in cells which were not activated.

Cell viability was confirmed by Trypan blue dye exclusion at the end of the incubation period.

Nitric oxide was determined by measurement of nitrate and nitrite in the cell supernatant. These anions are the stable end-products of reactions of NO in solution. Such reactions may or may not be catalysed in biological systems. The sum of nitrite and nitrate concentrations gives the total NO production. Nitrite was determined using the Griess reaction in which nitrite reacts with 1% sulphanilamide in 5% H_3PO_4/0.1% naphthylethylenediamine dihydrochloride to form a chromophore absorbing at 540nm. Nitrate was determined by reducing nitrate to nitrite with a bacterial nitrate reductase from *Pseudomonas oleovorans* and then measuring nitrite with the Griess reaction. In the absence of test compounds nitrite concentration plus nitrate concentration is equal to total nitric oxide production. The effect of test compounds on available nitric oxide (measured as nitrite + nitrate) was determined. The reduction in available nitric oxide compared with the control level may be taken as an indication of the degree of binding of NO by the test compounds.

In the *ex vivo* tests, segments of rat tail artery (0.8-1.5cm) were dissected free from normotensive adult Wistar rats. The arteries were internally perfused with Krebs solution (mM: NaCl 118, KCl 4.7, $NaHCO_3$ 25, NaH_2PO_4 1.15, $CaCl_2$ 2.5, $MgCl_2$ 1.1, glucose 5.6 and gassed with 95% O_2/5% CO_2 to maintain a pH of 7.4) in a constant flow perfusion apparatus. A differential pressure transducer located upstream of the vessel detected changes in back

pressure. The rat tail artery preparation was pre-contracted with 6.5μM phenylephrine to give a physiologically normal pressure of 100-120mm Hg. The pre-contracted vessels were then perfused with the test compound. The arteries were perfused with Krebs solution between applications of test compound to wash out the test compound.

Pressure changes in the system served to indicate artery vasoconstriction. The vasoconstriction is a direct result of the removal of endogenous nitric oxide (edrf) from the endothelial cells of the rat tail artery.

RESULTS

The results of the *in vitro*, *in vitro* cell culture and *ex-vivo* tests were as follows:

IN VITRO TESTS

EXAMPLE 1 : $K[Ru(Hedta)Cl]2H_2O$

A pressure decrease indicated binding of NO to the metal compound. This is illustrated in Figure 1.

The IR spectrum showed a peak at $1897cm^{-1}$, indicating the presence of a Ru-NO bond.

EXAMPLE 2 : [Ru(H_2edta)(acac)]

The IR spectrum showed a peak at 1896cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 3 : K[Ru(hedtra)Cl]H_2O

A pressure decrease indicated binding of NO to the metal compound. This is illustrated in Figure 1.

The IR spectrum showed a peak at 1889cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 4 : K[Ru(dipic)$_2$]H_2O

A pressure decrease indicated binding of NO to the metal compound. This is illustrated in Figure 1.

The IR spectrum showed a peak at 1915cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 5 : $(H_2pic)[RuCl_2(pic)_2](Hpic)H_2O$

The IR spectrum showed a peak at 1888cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 6 : $K[Ru(H_2edta)Cl_2]H_2O$

A pressure decrease indicated binding of NO to the metal compound. This is illustrated in Figure 1.

The IR spectrum showed a peak at 1896cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 7 : $K[Ru(Hnta)_2]1/2H_2O$

A pressure decrease indicated binding of NO to the metal compound. This is illustrated in Figure 1.

The IR spectrum showed a peak at 1889cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 8 : $K[Ru(H_2dtpa)Cl]H_2O$

A pressure decrease indicated binding of NO to the metal compound. This is illustrated in Figure 1.

The IR spectrum showed a peak at 1905cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 9 : $[Ru_3O(lac)_6](lac)$

The IR spectrum showed a peak at 1884cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 10 : $[Ru_3O(OAc)_6](OAc)$

The IR spectrum showed a peak at 1877cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 11 : $[Ru_2(OAc)_4]NO_3$

The IR spectrum showed a peak at 1891cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 12 : $[Ru(OCOEt)_4]NO_3$

The IR spectrum showed a peak at 1891cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 13 : $K_3[Ru(ox)_3]$

The IR spectrum showed a peak at 1889cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 14 : $[Ru_2(OAc)_4]Cl$

The IR spectrum showed a peak at 1895cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 15 : $[Ru(NH_3)_5Cl]Cl_2$

The IR spectrum showed two peaks at 1909cm^{-1} and 1928cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 16 : $[Ru(en)_3]I_3$

The IR spectrum showed a peak at 1906cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 17 : $K[RuCl_4(phen)]H_2O$

The IR spectrum showed a peak at 1904cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 18 : $[Ru(cyclam)Cl_2]Cl$

The IR spectrum showed a peak at 1895cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 19 : $K[RuCl_4(bipy)]$

The IR spectrum showed a peak at 1885cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 20 : $[RuCl_3(dmso)_2(NH_3)]$

The IR spectrum showed a peak at 1889cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 21 : $[Ru(NH_3)_6]Cl_3$

The IR spectrum showed a peak at 1910cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 22 : cis-[$RuCl_2(dmso)_4$]

The IR spectrum showed a peak at 1881cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 23 : cis-[$RuCl_2(dmso)_3(NH_3)$]

The IR spectrum showed a peak at 1893cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 24 : [$RuCl_3(dmso)_3$]

The IR spectrum showed a peak at 1880cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 25 : [$Ru(mtc)_3$]

The IR spectrum showed a peak at 1862cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 26 : [$Ru(maltol)_3$]

The IR spectrum showed a peak at 1866cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 27 : $[Ru(acac)_2(MeCN)_2]CF_3SO_3$

The IR spectrum showed a peak at 1899cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 28 : $K_2[RuCl_5(H_2O)]$

The IR spectrum showed a peak at 1903cm^{-1}, indicating the presence of a Ru-NO bond.

EXAMPLE 29 : $[Os(ox)(bipy)_2]H_2O$

The IR spectrum showed a peak at 1894cm^{-1}, indicating the presence of a Os-NO bond.

IN VITRO CELL CULTURE TESTS

Results are shown in Table 1 and Figure 2.

EXAMPLE 1: $K[Ru(Hedta)Cl]2H_2O$

Available nitric oxide was reduced in a dose-dependent fashion with a maximum reduction of 75% at a concentration of 100μM.

EXAMPLE 2: [$Ru(H_2edta)(acac)$]

Available nitric oxide was reduced by 82% at 100μM test compound.

EXAMPLE 3: $K[Ru(Hedtra)Cl]H_2O$

Available nitric oxide was reduced by 42% at 100μM.

EXAMPLE 6: $K[Ru(H_2edta)Cl_2]H_2O$

Available nitric oxide was reduced by 77% at 100μM test compound.

EXMAPLE 14: $[Ru_2(OAc)_4]Cl$

Available nitric oxide was reduced by 47% at 100μM.

EXAMPLE 15: $[Ru(NH_3)_5Cl]Cl_2$

Available nitric oxide was reduced by 86% at 100μM test compound.

EXAMPLE 26: $[Ru(maltolato)_3]$

Available nitric oxide was reduced by 71% at 100μM.

TABLE 1

			% Decrease of Available Nitric Oxide
	Example 1	25µM	12
		50µM	23
		100µM	75
	Example 2	100µM	82
	Example 3	100µM	42
	Example 6	100µM	77
	Example 14	100µM	47
	Example 15	100µM	86
	Example 26	100µM	71

EX-VIVO TESTS

EXAMPLE 2

Application of test compound resulted in a dose-dependent vasoconstriction at 10µM and 100µM. This effect was reversible by washout with Krebs solution.

EXAMPLE 3

Application of test compound resulted in a dose-dependent vasoconstriction at 10μM and 100μM. This effect was reversible by washout with Krebs solution.

EXAMPLE 14

Application of test compound resulted in a dose-dependent vasoconstriction at 10μM and 100μM. This effect was reversible by washout with Krebs solution.

EXAMPLE 15

Application of test compound resulted in a dose-dependent vasoconstriction at 10μM and 100μM. This effect was reversible by washout with Krebs solution.

EXAMPLE 26

Application of test compound resulted in a dose-dependent vasoconstriction at 10μM and 100μM and 1000μM. This effect was reversible by washout with Krebs solution.

TABLE 2

		% Vasoconstriction
Example 2	10μM	20
	100μM	69
Example 3	10μM	17
	100μM	59
Example 14	10μM	11
	100μM	40
Example 15	10μM	16
	100μM	86
Example 26	10μM	10
	100μM	18
	1000μM	25

CLAIMS

1. The use of a neutral, anionic or cationic metal complex having at least one site for coordination with NO, of formula

$$[M_a(X_bL)_cY_dZ_e]^{n\pm} \qquad \text{formula I,}$$

in the manufacture of a medicament for the attenuation of NO levels where NO in implicated in disease,

where:
- M is a metal ion or a mixture of metal ions;
- X is a cation or a mixture of cations;
- L is a ligand, or mixture of ligands each containing at least two different donor atoms selected from the elements of Group IV, Group V or Group VI of the Periodic Table;
- Y is a ligand, or a mixture of the same or different ligands each containing at least one donor atom or more than one donor atom, which donor atom is selected from the elements of Group IV, Group V or Group VI of the Periodic Table;

and
- Z is a halide or pseudohalide ion or a mixture of halide ions and pseudohalide ions;

a = 1-3; b = 0-12; c = 0-18; d = 0-18; e = 0-18; and n = 0-10;

provided that at least one of c, d and e is 1 or more;

and where c is 0; b is also 0;

and where a is 1; c, d and e are not greater than 9;

and where a is 2; c, d and e are not greater than 12.

2. The use of a complex of formula I in the manufacture of a medicament for the treatment of NO-overproduction related disease.

3. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to claim 1, wherein M is a first, second or third row transition metal ion.

4. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to claim 1, wherein M is an Rh, Ru, Os, Mn, Co, Cr or Re ion.

5. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to claim 4, wherein M is an Rh, Ru or Os ion.

6. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to claim 5, wherein M is in oxidation state III.

7. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to any of claims 1-6, wherein X is a mono-, di- or tri-valent cation.

8. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to claim 7, wherein X is H^+, K^+, Na^+, NH_4^+ or Ca^{2+}.

9. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to any of claims 1-8, wherein L is a ligand containing both nitrogen and oxygen donor atoms.

10. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to claim 9, wherein L is edda, edta, nta, dipic, pic, dtpa, hedtra, tedta or dtedta.

11. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to any of claims 1-10, wherein Y is a ligand containing at least one of N, O, S, C or P donor groups.

12. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to claim 11, wherein said N donor group is ammine, amine, amide, nitrile or nitride or derivatives thereof.

13. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to claim 11, wherein said O donor group is carboxylic acid, ester or derivatives thereof, water, oxide, sulphoxide, hydroxide, acetate, lactate, propionate, oxalate or maltolate.

14. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to claim 11, wherein said S donor group is sulphoxide, dialkylsulphide, dialkylcarbamate, dithiocarbamate, or dithiophosphate.

15. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to claim 11, wherein said C donor group is carbon monoxide or isocyanide.

16. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to claim 11, wherein said P donor group is trialkylphosphine.

17. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to any preceding claim, wherein Z is a halide.

18. The use of a complex of formula I in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease, according to claim 17, wherein Z is chloride.

19. The use of an optionally hydrated ruthenium complex of formula

$$[Ru(H_{0-6}L'')_{1-3}Y_{0-2}Cl_{1-4}]^{(0-4)\pm} \qquad \text{formula II,}$$

where L" is an amide or ester or derivative thereof, or a polydentate aminocarboxylate ligand, for example edda, tropolone edta, nta, dipic, pic, dtpa, hedtra, tedta or dtedta or a mixture of any of these, and Y is as defined above and may for example be selected from acetylacetone (acac), a β-diketonate, water, dimethylsulphoxide (dmso), carboxylate, bidentate carboxylate, catechol, kojic acid, maltol, hydroxide, tropolone, malonic acid, oxalic acid, 2,3-dihydroxynaphthalene, squaric acid, acetate, a sulphate and a glycolate in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease.

20. A pharmaceutical composition containing an optionally hydrated complex of formula

$$[M_{1-3}Y_{1-18}Cl_{0-18}]^{(o-6)\pm} \qquad \text{formula III}$$

wherein Y is a sulphur donor ligand.

21. The use of an optionally hydrated complex of ruthenium of formula

$$[M'''_{1-3}Y'''_{1-18}Cl_{0-18}]^{(0-6)\pm} \qquad \text{formula III}$$

where M''' is ruthenium and Y''' is an oxygen-donor ligand such as acetate, lactate, water, oxide, propionate, oxalate or maltolate or a combination of these, in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease.

22. The use of an optionally hydrated complex of ruthenium of formula

$$[RuY^{IV}_{1-9}Cl_{1-9}]^{(0-4)\pm} \qquad \text{formula IV}$$

where Y^{IV} is a nitrogen-donor ligand such as ammine, ethylenediamine (en), pyridine (py), 1,10-phenanthroline (phen), 2,2'-bipyridine (bipy), 1,4,8,11- tetraaza-cyclotetradecane (cyclam), 2,3,7,8,12,13,17,18-octaethylporphyrin (oep) or a

combination of these, in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease.

23. The use of an optionally hydrated complex of ruthenium or osmium of general formula

$$[M_{1-3}Y_{1-18}Cl_{0-18}]^{(0-6)\pm} \qquad \text{formula V}$$

where Y is a combination according to claims 19, 20, 21, or 22 in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease.

24. The use of an optionally hydrated complex of general formula

$$[M_{1-3}Y_{1-18}Cl_{0-18}]^{(0-6)\pm}$$

where Y is a sulphur donor ligand, in the manufacture of a medicament for the attenuation of NO levels where NO is implicated in disease.

25. A pharmaceutical composition containing an optionally hydrated ruthenium complex as described in claim 19.

26. A pharmaceutical composition containing an optionally hydrated complex of ruthenium as described in claim 21, wherein Y''' is an oxygen donor ligand selected from the group acetate, lactate, oxide, propionate and maltolate.

27. A pharmaceutical composition containing an optionally hydrated complex of ruthenium as described in claim 22, wherein Y^{IV} is a nitrogen donor ligand selected from the group en, py, phen, bipy, cyclam or oep.

28. A pharmaceutical composition containing an optionally hydrated complex of formula

$[Os(ox)(bipy)_2]$

29. A pharmaceutical composition containing an optionally hydrated complex of formula

$[Ru(acac)_2(MeCN)_2]^+$

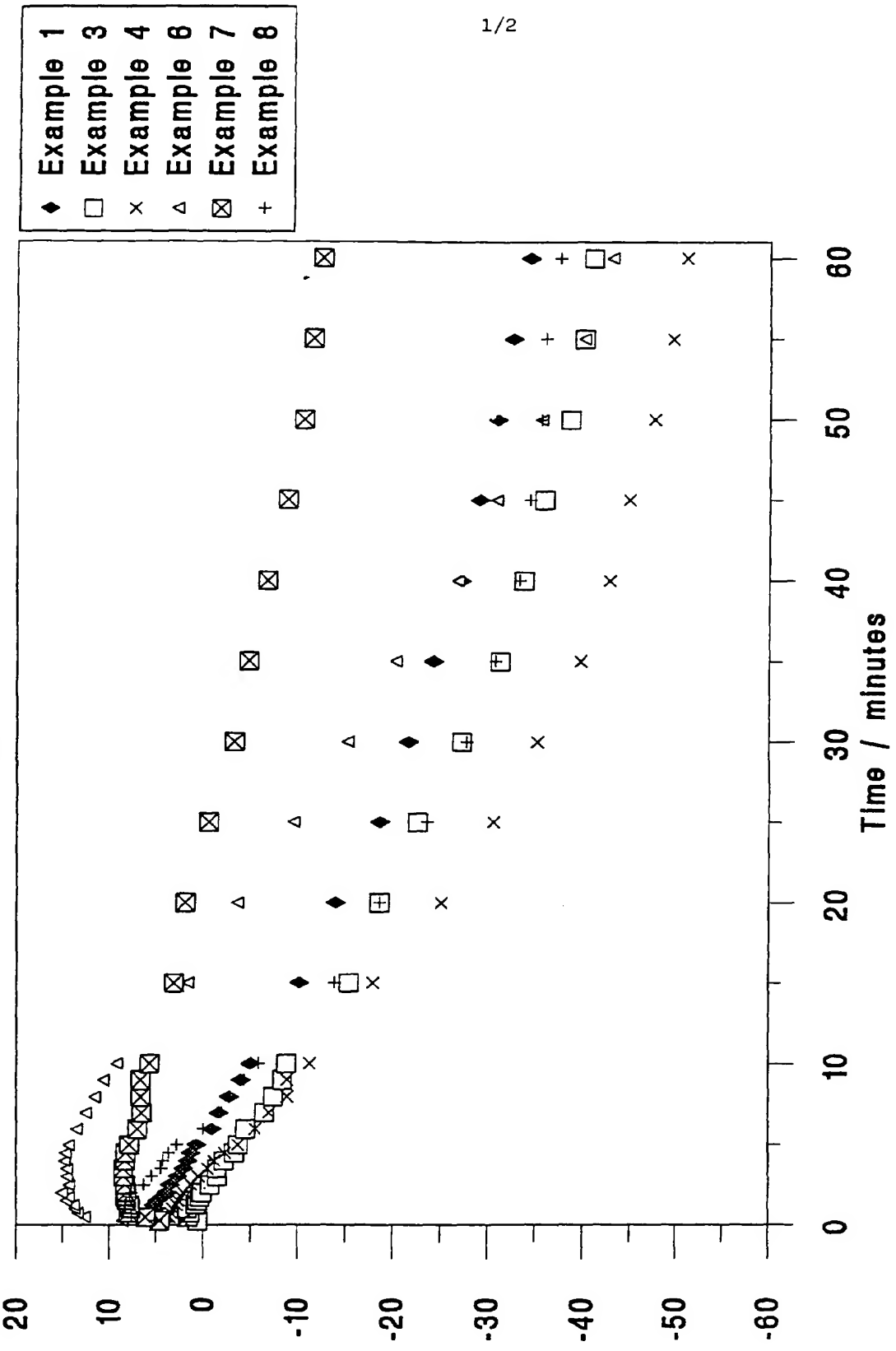
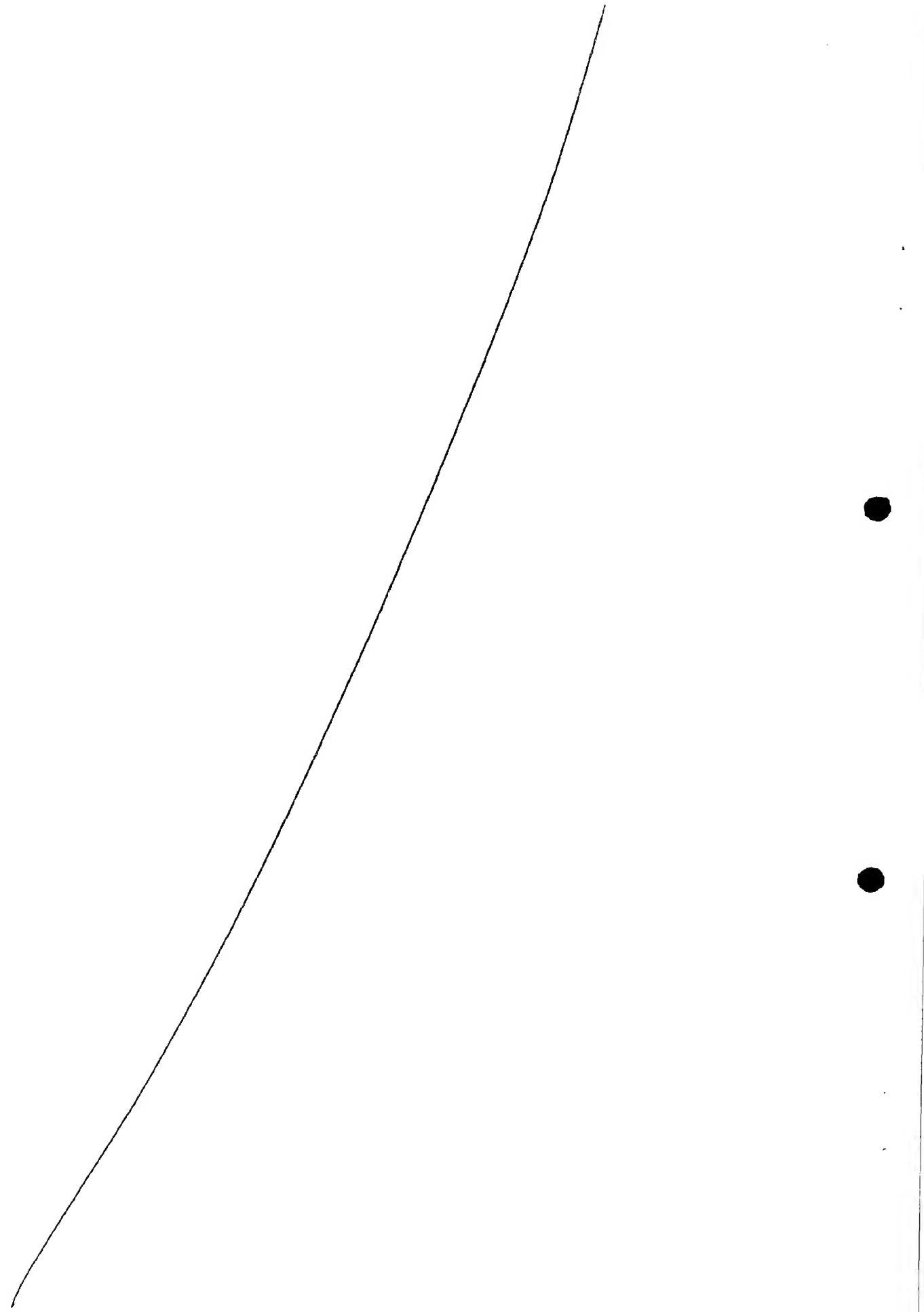

Example 1
Example 3
Example 4
Example 6
Example 7
Example 8
Time / minutes
0
10
20
30
40
50
60
Pressure Change
20
10
0
-10
-20
-30
-40
-50
-60

Figure 1



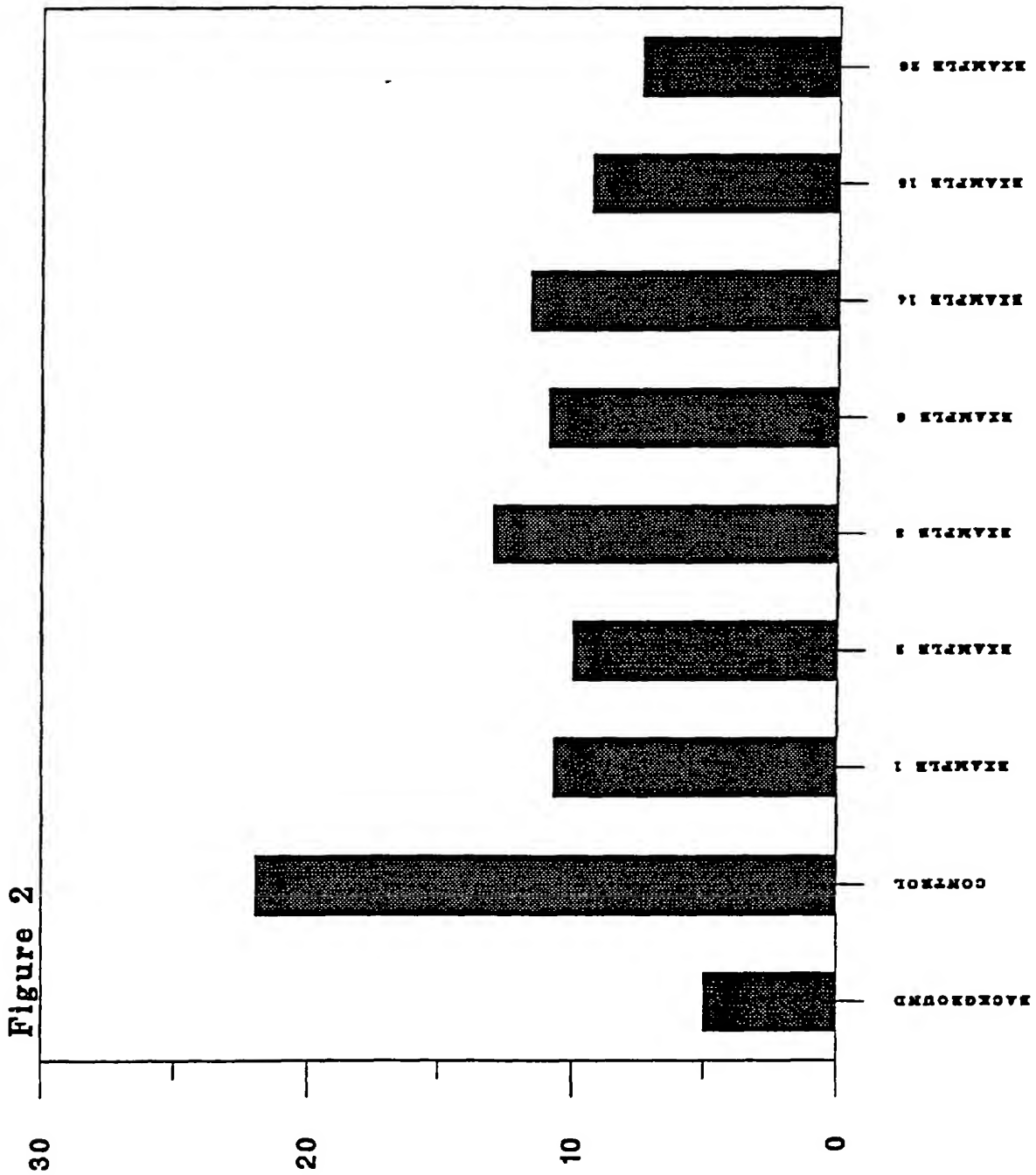

BACKGROUND
CONTROL
EXAMPLE 1
EXAMPLE 3
EXAMPLE 5
EXAMPLE 6
EXAMPLE 14
EXAMPLE 15
EXAMPLE 16
0
10
20
30
AVAILABLE NITRIC OXIDE (micromoles/l)

Figure 2